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Nomenclatural status and morphological notes on *Tubifera applanata* sp. nov. (*Myxomycetes*)

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Abstract — *Tubifera applanata* sp. nov. is proposed to validate "*Tubulifera applanata*" nom. inval. The species diagnosis and some notes on its morphology are provided. At 0.40–0.65 mm diam., individual sporothecae are somewhat larger than in *T. ferruginosa* and *T. microsperma* and smaller than those in *T. casparyi*. Circular ornamentations on the inner peridial surface in *T. applanata* are larger than previously noted, reaching a size up to 2.9 µm.

Key words — morphometry, SEM, species delimitation

Introduction

In 2001, C. Lado stated that the invalid generic name "*Tubulifera*" O.F. Müll. 1775 had been validated as *Tubulifera* O.F. Müll. ex Jacq. 1779, a name with priority over *Tubifera* J.F. Gmel. 1792 (Lado 2001). On this understanding, Leontyev & Fefelov (2009) proposed a new species, *Tubulifera applanata*.

Subsequently, Lado (2011) indicated that, contrary to previous opinion, Jacquin's publication failed to validate the genus "*Tubulifera.*" This means that all species names published in "*Tubulifera,*" including "*Tubulifera applanata,*" are also invalid (ICBN [Vienna Code] Article 43.1). We therefore propose a validation of "*Tubulifera applanata*" as *Tubifera applanata* sp. nov. and provide additional morphological data.

Materials & methods

A total of 13 specimens of *Tubifera applanata* were examined (CWU MR 001, 038a, 038b, 039, 054, 058, 074, 077, 121, 122, 123, 125, 126). Additionally, 8 specimens of *T. ferruginosa* (Batsch) J.F. Gmel. (CWU MR 006, 023, 012, 091, 095, 103, 113, 114), 4 of *T. microsperma* (Berk. & M.A. Curtis) G.W. Martin (CWU MR 008, 041, 049, 153), and 3 of *T. casparyi* (Rostaf.) T. Macbr. (CWU MR 049, 181, 183) were used for comparison. All the specimens were collected during 2003–2010 in the East Forest-Steppe region

of Ukraine (Gomolsha Forests National Nature Park, Kharkiv region). Material was deposited in the herbarium of the V.N. Karasin National University of Kharkiv (CWU), the subsection of Reticulariaceae (MR).

The specimens were examined using a stereoscopic microscope MBS-9 and scanning electron microscope (SEM) Jeol JSM-6060 (30 kV, gold evaporation).

The diameters of intact closed sporothecae were measured from above by a micrometer. After drawing an imaginary line corresponding to the largest fruit body diameter on the surface of the pseudoaethalium, we measured 30 sporothecae along this line from the edge of the pseudoaethalium. The diameters of annular ornamentations on the inner peridial surface were measured using the SEM scale.

Means, standard deviations, and standard errors were calculated using Stat Soft Statistica 8.0 and Microsoft Excel 2003 software to describe the morphological variety.

Results & discussion

Tubifera applanata Leontyev & Fefelov, **sp. nov.** Fig. 1c–d

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"*Tubulifera applanata*" Leontyev & Fefelov, Bol. Soc. Mycol. Madrid 33: 119. 2009

Differs from *Tubifera ferruginosa* by its large pseudoaethalia, flat sporothecal tips, annular embossments on the internal peridial surface, and salmon color of young fructifications.

Type: Ukraine, Kharkiv region, Gomolsha Forests National Nature Park, Zadinetske forestry, edge of pine forest near 'Bile Ozero' Camp (49°36′50.69″N 36°21′06.15″E), on the bark of fallen trunk of *Pinus sylvestris* L., 14.07.2003, leg. D.V. Leontyev (**Holotype**, CWU MR 039).

Etymology: *applanatus* (Lat.) = flat; referring to the flattened sporothecal tips.

Pseudoaethalium large, (12–)23–37(–73) mm long, (7–)16.5–27.5(–38) mm wide, 4–6 mm in height, oval or irregular as seen from above, flat pulvinate, rust-brown. Sporothecae straight, clearly prismatic from mutual pressure. Tips of sporothecae 0.4–0.65 mm diam., flat or slightly convex, 5–6 angular or almost circular, equal in height, forming a smooth surface of pseudoaethalium. Peridium opaque, light brown, lateral walls of the sporothecae often plicate. Internal surface of peridium covered with annular embossments (0.4–)1.5–2.9 μm diam., quite distant from each other. Columella absent or not reaching the top of the sporothecae, thick, with color of spore mass. Pseudocapillitium absent or scanty, filiform, appearing as occasionally branched strings. Hypothallus spongy, poorly developed or absent. Spores in mass rust-brown, brownish by transmitted light, 4.9–6.3(–7.5) μm diam., banded reticulate. Plasmodium salmon or rusty-cream. In coniferous forests, on bark, litter and slightly decomposed wood.

Notes on the morphology

Tubifera applanata is characterized by the prismatic shape of the individual sporothecae, their flat hexagonal tips, ring-like ornamentations on the inner peridial surface, and the salmon color of young fructifications. Although we

first noted that quantitative parameters such as pseudoaethalium size and spore diameter distinguished *T. applanata* from related species (Leontyev & Fefelov 2009), we did not realize that individual sporothecal diameters might also serve as a diagnostic criterion. Moreover, there were no data referring to the diameter of sporothecae in other *Tubifera* species (Nannenga-Bremekamp 1991, Neubert et al. 1993, Ing 1999).

The study of sporothecae size in *Tubifera applanata, T. ferruginosa, T. microsperma,* and *T. casparyi* has shown that all species have significant differences ($p<0.01$) from one another. Measurements show that individual sporothecae are somewhat larger in *T. applanata* than in *T. ferruginosa* and *T. microsperma* and smaller than those in *T. casparyi* (Table 1).

Table 1. Sporothecal diameters in four *Tubifera* species.

	T. applanata	*T. ferruginosa*	*T. microsperma*	*T. casparyi*
Range of specimen means (μm)	464–633	322–474	330–398	512–692
Mean (μm)	515	398	374	574
Standard deviation [sd] (μm)	116.0	95.2	67.6	98.2
Mean ± sd (μm)	399–631	302–493	306–442	476–672
Rounded variation, for use in species descriptions (mm)	0.4–0.65	0.3–0.5	0.3–0.45	0.5–0.7

Although the difference in sporothecae size between *T. applanata* and *T. ferruginosa* is quite small (0.1–0.2 mm), it can be seen with the naked eye. We propose to use this criterion as an additional character in descriptions of *Tubifera* species.

Peridium ultrastructure

Nelson et al. (1982) introduced a new diagnostic criterion into *Tubifera* taxonomy — the type of inner peridial ornamentation, noting that in *T. microsperma* the inner surface of the peridium is ornamented by so-called 'rimmed craters.' In Ukrainian specimens of *T. microsperma*, the ornamentation agrees with this description (Fig. 1A–B).

In 2009 the SEM-study of *T. applanata* showed 'peridial rings,' similar to those of *T. microsperma* but much lower and more distant from each other (Leontyev & Fefelov 2009). Both 'peridial rings' and 'rimmed craters' appear to be remnants of channels used by the plasmodium for active water evacuation (R. McHugh, pers. comm.).

We previously indicated the outer 'peridial ring' diameter in *T. applanata* as 0.4–0.9 μm (Leontyev & Fefelov 2009). Improved SEM techniques demonstrate that in the type specimen, these structures reach 1.6–2.9 μm diam. (Fig. 1C–D), significantly larger than similar structures in *T. microsperma*. In *T. applanata,* rings < 1 μm diam. are usually less prominent than the larger ones.

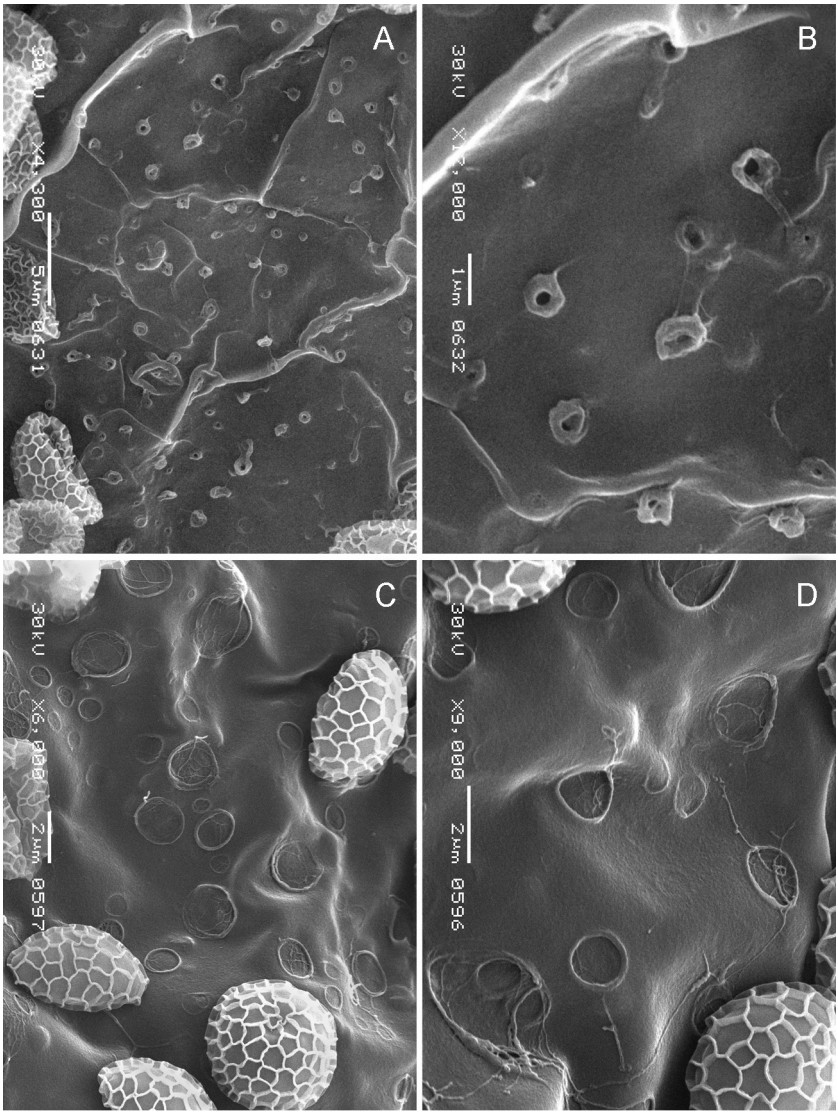

Figure 1. A–B: *Tubifera microsperma* [CWU MR 135]—ornamentation of the inner peridium surface illustrating 'rimmed craters.' C–D: *Tubifera applanata* [CWU MR 039, holotype]—ornamentation of the inner peridium surface illustrating 'peridial rings' and intricate filaments.

This study has shown that the peridial surface of *T. applanata* is ornamented by thin intricate filaments, appearing to be composed of dried slime (Fig. 1D). Such filaments are especially abundant in the bottom of the 'peridial rings.'

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